

BRIEF COMMUNICATION

Serum α -Tocopherol and γ -Tocopherol in Relation to Prostate Cancer Risk in a Prospective Study

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The Alpha-Tocopherol, Beta-Carotene Cancer Prevention (ATBC) Study demonstrated a 32% reduction in prostate cancer incidence in response to daily α -tocopherol supplementation. We examined baseline serum concentrations of α -tocopherol and γ -tocopherol to compare their respective associations with prostate cancer risk. From the ATBC Study cohort of 29 133 Finnish men, 50–69 years old, we randomly selected 100 incident prostate cancer case patients and matched 200 control subjects. Odds ratios and 95% confidence intervals (CIs) were estimated for the serum tocopherols (measured by high-performance liquid chromatography) using logistic regression models. All *P* values were two-sided. Odds ratios for the highest versus the lowest tertiles were 0.49 (95% CI = 0.24 to 1.01, $P_{\text{trend}} = .05$) for α -tocopherol and 0.57 (95% CI = 0.31 to 1.06, $P_{\text{trend}} = .08$) for γ -tocopherol. Further analyses indicated that the association of high serum tocopherols with low prostate cancer risk was stronger in the α -tocopherol-supplemented group than in those not receiving α -tocopherol. Participants with higher circulating concentrations of the major vitamin E fractions, α -tocopherol and γ -tocopherol, had similarly lower prostate cancer risk. [J Natl Cancer Inst 2005; 97:396–9]

Vitamin E occurs naturally as four tocopherols and four tocotrienols. γ -Tocopherol, the most prevalent form of vitamin E in

the typical U.S. diet (1–3), has received increasing attention recently (2). α -Tocopherol is the predominant form of vitamin E in plasma, regardless of dietary intake, due to preferential binding by the hepatic α -tocopherol transfer protein (1–3).

α -Tocopherol supplementation reduced prostate cancer incidence by 32% (95% confidence interval [CI] = –47% to –12%) in the Alpha-Tocopherol, Beta-Carotene Cancer Prevention (ATBC) Study (4). Observational data regarding serum α -tocopherol and prostate cancer risk have been mixed, however (5–15), and few studies measured γ -tocopherol (7–11). We therefore conducted a nested case–control study within the ATBC Study cohort to compare the prostate cancer risk associations of serum α -tocopherol and γ -tocopherol.

The ATBC Study included 29 133 male smokers, aged 50–69 years, recruited from southwestern Finland from 1985 to 1988. Subjects were provided α -tocopherol and/or β -carotene supplements or placebo for 5–8 years. The study was approved by the institutional review boards of the National Cancer Institute and the National Public Health Institute of Finland, and written informed consent was obtained from all participants (16).

Case patients (*n* = 100) were randomly selected from among 246 incident prostate cancer patients ascertained through the Finnish Cancer Registry, based on the abstracted medical records, and diagnosed through April 30, 1993. The time from baseline to prostate cancer diagnosis ranged from 2.2 to 7.9 years (median, 6.1 years). Control subjects (*n* = 200) were alive and free of prostate cancer at the time of case patient diagnosis and were individually matched to case patients by age (within 5 years), intervention group, and date of baseline serum collection (within 15 days).

At baseline, participants completed risk factor and dietary questionnaires (17) and provided fasting serum samples (stored at –70 °C). Serum α -tocopherol and γ -tocopherol concentrations were determined by reverse-phase high-performance liquid chromatography, as previously described (18). Case patients and control subjects were assayed consecutively within batches along with blinded quality-control samples (*n* = 32). Coefficients of variation were 2.6% (within-batch) and 3.3% (between-batches) for α -tocopherol and 6.2% and

4.7%, respectively, for γ -tocopherol. Serum β -carotene and cholesterol were previously measured (16,19). Odds ratios (ORs) and 95% CIs were estimated using conditional logistic regression models, adjusted for serum cholesterol. Age at randomization, body mass index, height, smoking, benign prostatic hyperplasia, physical activity, urban residence, education, and marital status were not confounders in our sample (i.e., each factor produced <10% change in tocopherol beta-coefficients). Effect modification was assessed through a cross-product term and by stratification. All *P* values were two-sided, and groups were considered statistically significantly different if *P* < .05.

Case patients had lower intake of total vitamin E but were otherwise comparable to control subjects (Table 1). In contrast to patterns in U.S. populations, α -tocopherol intake exceeded γ -tocopherol intake in these Finnish men. Serum α -tocopherol and γ -tocopherol were highly correlated (all *P* < .01) with each other (Spearman *r* = 0.51), with intakes of total vitamin E (*r* = 0.22 and 0.24, respectively), α -tocopherol (*r* = 0.20 and 0.22), γ -tocopherol (*r* = 0.20 and 0.33), and serum cholesterol (*r* = 0.61 and 0.22).

Men with higher circulating levels of α -tocopherol and γ -tocopherol had lower prostate cancer risk, and the relationships were concentration dependent and demonstrated borderline statistical significance for α -tocopherol (Table 2). When both tocopherols were included in the model, odds ratios were attenuated (for α -tocopherol, OR = 0.58, 95% CI = 0.26 to 1.26; for γ -tocopherol, OR = 0.68, 95% CI = 0.35 to 1.31 for the highest versus the lowest tertiles).

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Table 1. Selected baseline characteristics of prostate cancer patients and control subjects, ATBC Study*

Characteristic	Mean (SD)	
	Case patients (n = 100)	Control subjects (n = 200)
Age, years	61.0 (4.7)	60.3 (4.4)
Height, cm	173.5 (6.2)	173.9 (6.0)
Weight, kg	79.3 (12.0)	78.4 (12.7)
Body mass index, kg/m ²	26.3 (3.4)	25.9 (3.7)
No. of cigarettes smoked per day	18.9 (7.4)	18.8 (8.1)
Years of smoking	39.9 (8.2)	39.2 (7.6)
Benign prostatic hyperplasia, %	10	6
Family history of prostate cancer, %†	4.7	3.8
Physical activity, % active‡	11	19
Use of supplement with vitamin E, %	11	10
Urban residence, %	50	48
Married, %	80	85
Education, % > elementary school	17	17
Daily dietary intake		
Total energy, kcal	2697 (687)	2874 (758)
Total vitamin E, mg α -tocopherol equivalents	11.6 (5.5)	13.0 (6.0)§
α -Tocopherol, mg	10.1 (5.4)	11.0 (5.0)
γ -Tocopherol, mg	7.7 (6.9)	9.2 (8.2)
Biomarker serum concentration		
α -Tocopherol, mg/dL	1.34 (0.34)	1.42 (0.39)
γ -Tocopherol, mg/dL	0.09 (0.04)	0.10 (0.04)
β -Carotene, μ g/mL	0.24 (0.32)	0.24 (0.19)
Cholesterol, mM	6.3 (1.2)	6.5 (1.4)
HDL cholesterol, mM	1.2 (0.3)	1.2 (0.3)

*ATBC = Alpha-Tocopherol, Beta-Carotene Cancer Prevention (cohort of male Finnish smokers, ages 50–69 years, recruited from 1985 to 1988), SD = standard deviation, HDL = high-density lipoprotein.

†Based on 43 case patients and 133 control subjects with available data.

‡Light or moderate activity at work and moderate or heavy activity at leisure.

§Wilcoxon $P = .04$ (two-sided).

The ratio of serum α -tocopherol to γ -tocopherol was not associated with risk. We jointly classified participants based on the median values of α -tocopherol and γ -tocopherol among the control subjects and observed the following odds ratios: 0.58, 0.66, and 0.55 for high α -tocopherol/low γ -tocopherol, low α -tocopherol/high γ -tocopherol, and high α -tocopherol/high γ -tocopherol compared with the group low in both tocopherols (all confidence intervals included 1.0). The associations for both tocopherols were stronger among study participants who received α -tocopherol supplementation and among those who received β -carotene supplementation than among those who did not (Table 2); however, none of the interactions was statistically significant. In addition, prostate cancer risk appeared lower among men in the highest tertiles of γ -tocopherol who had below-median vitamin E intake than among those with above-median intake and lower among those who reported use of any vitamin supplements than among those who did not (data not shown; interactions not statistically significant).

Five previous prospective studies examined both serum α -tocopherol and γ -tocopherol in relation to prostate

cancer risk (7–11). All but one (11) showed inverse associations for higher serum α -tocopherol concentrations (odds ratios ranging from 0.58 to 0.78 for highest versus lowest quantiles), the association was statistically significant in one study ($P = .04$) (10), and another found the association only among those with advanced disease (and slightly stronger still among smokers with advanced disease) (7). Only one study (two analyses from the Washington County, Maryland, “CLUE II” cohort) found an inverse association for serum γ -tocopherol (8,9). This study reported higher median concentrations of γ -tocopherol (0.28–0.29 mg/dL) than did the null studies [0.15–0.24 mg/dL (7,9–11)], and risk reduction was limited to the highest quintile (i.e., >0.41 mg/dL) (8,9). In the present investigation, participants had much lower γ -tocopherol concentrations (median = 0.10 mg/dL), yet we observed an inverse dose–response association with prostate cancer. In addition, the ratio of serum α -tocopherol to γ -tocopherol is highest in the ATBC Study and lowest in CLUE II, so neither a threshold effect nor the ratio of the two tocopherols appears to explain the relationship between γ -tocopherol and prostate cancer.

Previous data suggest that α -tocopherol supplementation decreases plasma and tissue γ -tocopherol concentrations (2,20). Although for this study we assayed presupplementation serum and cannot address this issue directly, our findings indicate an inverse association for baseline serum γ -tocopherol and prostate cancer risk, which was stronger among the men who were supplemented with 50 mg of α -tocopherol daily during the trial. Our previous cohort analysis showed a pattern for dietary γ -tocopherol similar to that observed here for serum γ -tocopherol (5). The data in the present study suggest that α -tocopherol supplementation did not negatively impact the γ -tocopherol/prostate cancer association and may have strengthened it. Interestingly, the tocopherol associations were also somewhat stronger in the β -carotene-supplemented group than in those who were not given β -carotene supplements. How supplementation with either α -tocopherol or β -carotene might modify the relationship between serum tocopherol concentrations and prostate cancer risk is a matter of speculation but may involve enhanced absorption, preferential carriage, membrane transport or function, or biochemical function of the tocopherols in those who received either supplement, or it could be due to chance. Of potential relevance is our observation that men in the lowest tertile of baseline serum α -tocopherol who were given the α -tocopherol supplement did not, on average, achieve serum α -tocopherol levels as high as the baseline level of men in the highest tertile. This suggests that, even with α -tocopherol supplementation, men whose usual levels were low may not have attained a threshold serum concentration.

The antioxidant activity of vitamin E may be particularly important to the observed associations because oxidative stress has been implicated in prostate carcinogenesis (21). α -Tocopherol has other important non-antioxidant functions as well, including enhancement of the immune response, modulation of gene expression, and inhibition of protein kinase C activity, cell proliferation, and cell adhesion (3,22). Recently, α -tocopheryl succinate was shown experimentally to inhibit prostate cancer cell growth through suppressed expression of the androgen receptor, prostate-specific antigen, and cell cycle regulatory proteins (23,24). We previously showed

Table 2. Adjusted ORs and 95% CIs of prostate cancer according to baseline serum α -tocopherol and γ -tocopherol and stratified by α -tocopherol or β -carotene supplementation group, ATBC Study*

Nutrient	No. of case patients/no. of control subjects			<i>P</i> _{trend}	<i>P</i> _{interaction}
	OR (and 95% CI) by tertile of serum tocopherol				
	1	2	3		
Serum α-tocopherol, mg/dL	44/66 1.00 (referent)	34/67 0.73 (0.40 to 1.33)	22/66 0.49 (0.24 to 1.01)	.05	
α-Tocopherol supplementation					
No	30/46 1.00 (referent)	20/41 0.78 (0.36 to 1.67)	15/42 0.63 (0.26 to 1.53)	.31	
Yes	14/20 1.00 (referent)	14/26 0.64 (0.23 to 1.76)	7/24 0.31 (0.09 to 1.10)	.07	.77
β-Carotene supplementation					
No	17/27 1.00 (referent)	14/27 0.84 (0.29 to 2.43)	9/25 0.67 (0.20 to 2.20)	.50	
Yes	27/39 1.00 (referent)	20/40 0.67 (0.32 to 1.41)	13/41 0.39 (0.15 to 0.99)	.05	.93
Serum γ-tocopherol, mg/dL	44/66 1.00 (referent)	32/67 0.74 (0.41 to 1.31)	24/66 0.57 (0.31 to 1.06)	.08	
α-Tocopherol supplementation					
No	26/48 1.00 (referent)	23/43 1.04 (0.52 to 2.09)	16/38 0.85 (0.40 to 1.83)	.69	
Yes	18/18 1.00 (referent)	9/24 0.31 (0.10 to 0.96)	8/28 0.24 (0.08 to 0.76)	.02	.11
β-Carotene supplementation					
No	18/29 1.00 (referent)	11/28 0.66 (0.25 to 1.73)	11/22 0.94 (0.36 to 2.47)	.88	
Yes	26/37 1.00 (referent)	21/39 0.78 (0.38 to 1.61)	13/44 0.42 (0.18 to 0.96)	.04	.34

*Case patients and control subjects were matched by age, trial intervention group, and date of baseline serum collection. ORs were adjusted for serum cholesterol. ATBC = Alpha-Tocopherol, Beta-Carotene Cancer Prevention. Cut points for tertiles of serum α -tocopherol are 1.260 and 1.578 mg/dL. Cut points for serum γ -tocopherol are 0.076 and 0.108 mg/dL. Cut points were based on equal distribution among control subjects. One control subject who did not have a serum cholesterol value was excluded from all analyses.

decreased androgen concentrations in response to α -tocopherol supplementation (25). γ -Tocopherol has some functions that differ from those of α -tocopherol, including, for example, protection against reactive nitrogen species (1,26) and selective inhibition of cyclooxygenase activity and prostaglandin E_2 synthesis (2,3).

In conclusion, higher prediagnostic circulating concentrations of the major vitamin E fraction, α -tocopherol, were associated with a substantially lower risk of prostate cancer; the association with γ -tocopherol was similar. The serum vitamin E concentrations measured in this study represent status prior to supplementation with α -tocopherol. In addition, the findings in this study were accentuated among men who received α -tocopherol supplementation, which may allay concerns regarding whether supplementation with α -tocopherol may negatively impact γ -tocopherol status in other prevention

trials such as the Selenium and Vitamin E Cancer Prevention Trial (SELECT) (27).

REFERENCES

- (1) Traber MG, Arai H. Molecular mechanisms of vitamin E transport. *Annu Rev Nutr* 1999;19:343–55.
- (2) Jiang Q, Christen S, Shigenaga MK, Ames BN. Gamma-tocopherol, the major form of vitamin E in the US diet, deserves more attention. *Am J Clin Nutr* 2001;74:714–22.
- (3) Brigelius-Flohe R, Kelly FJ, Salonen JT, Neuzil J, Zingg JM, Azzi A. The European perspective on vitamin E: current knowledge and future research. *Am J Clin Nutr* 2002;76:703–16.
- (4) Heinonen OP, Albanes D, Virtamo J, Taylor PR, Huttunen JK, Hartman AM, et al. Prostate cancer and supplementation with alpha-tocopherol and beta-carotene: incidence and mortality in a controlled trial. *J Natl Cancer Inst* 1998;90:440–6.
- (5) Hartman TJ, Albanes D, Pietinen P, Hartman AM, Rautalahti M, Tangrea JA, et al. The association between baseline vitamin E,

selenium, and prostate cancer in the alpha-tocopherol, beta-carotene cancer prevention study. *Cancer Epidemiol Biomarkers Prev* 1998;7:335–40.

- (6) Eichholzer M, Stahelin HB, Gey KF, Ludin E, Bernasconi F. Prediction of male cancer mortality by plasma levels of interacting vitamins: 17-year follow-up of the prospective Basel study. *Int J Cancer* 1996;66:145–50.
- (7) Gann PH, Ma J, Giovannucci E, Willett W, Sacks FM, Hennekens CH, et al. Lower prostate cancer risk in men with elevated plasma lycopene levels: results of a prospective analysis. *Cancer Res* 1999;59:1225–30.
- (8) Helzlsouer KJ, Huang HY, Alberg AJ, Hoffman S, Burke A, Norkus EP, et al. Association between alpha-tocopherol, gamma-tocopherol, selenium, and subsequent prostate cancer. *J Natl Cancer Inst* 2000;92:2018–23.
- (9) Huang HY, Alberg AJ, Norkus EP, Hoffman SC, Comstock GW, Helzlsouer KJ. Prospective study of antioxidant micronutrients in the blood and the risk of developing prostate cancer. *Am J Epidemiol* 2003;157:335–44.
- (10) Goodman GE, Schaffer S, Omenn GS, Chen C, King I. The association between lung and prostate cancer risk, and serum micronutrients: results and lessons learned from beta-carotene and retinol efficacy trial. *Cancer Epidemiol Biomarkers Prev* 2003;12:518–26.
- (11) Nomura AM, Stemmermann GN, Lee J, Craft NE. Serum micronutrients and prostate cancer in Japanese Americans in Hawaii. *Cancer Epidemiol Biomarkers Prev* 1997;6:487–91.
- (12) Hayes RB, Bogdanovic JF, Schroeder FH, De Bruijn A, Raatgever JW, Van der Maas PJ, et al. Serum retinol and prostate cancer. *Cancer* 1988;62:2021–6.
- (13) Knekt P, Aromaa A, Maatela J, Aaran RK, Nikkari T, Hakama M, et al. Serum vitamin E and risk of cancer among Finnish men during a 10-year follow-up. *Am J Epidemiol* 1988;127:28–41.
- (14) Hsing AW, Comstock GW, Abbey H, Polk BF. Serologic precursors of cancer. Retinol, carotenoids, and tocopherol and risk of prostate cancer. *J Natl Cancer Inst* 1990;82:941–6.
- (15) Comstock GW, Helzlsouer KJ, Bush TL. Prediagnostic serum levels of carotenoids and vitamin E as related to subsequent cancer in Washington County, Maryland. *Am J Clin Nutr* 1991;53:260S–264S.
- (16) The ATBC Cancer Prevention Study Group. The alpha-tocopherol, beta-carotene lung cancer prevention study: design, methods, participant characteristics, and compliance. The ATBC Cancer Prevention Study Group. *Ann Epidemiol* 1994;4:1–10.
- (17) Pietinen P, Hartman AM, Haapa E, Rasanen L, Haapakoski J, Palmgren J, et al. Reproducibility and validity of dietary assessment instruments. I. A self-administered food use questionnaire with a portion size picture booklet. *Am J Epidemiol* 1988;128:655–66.
- (18) Zhang C, Williams MA, Sanchez SE, King IB, Ware-Jauregui S, Larrabure G, et al. Plasma concentrations of carotenoids, retinol, and tocopherols in preeclamptic and normotensive pregnant women. *Am J Epidemiol* 2001;153:572–80.

- (19) Leppala JM, Virtamo J, Fogelholm R, Albanes D, Heinonen OP. Different risk factors for different stroke subtypes: association of blood pressure, cholesterol, and antioxidants. *Stroke* 1999;30:2535–40.
- (20) Handelman GJ, Machlin LJ, Fitch K, Weiter JJ, Dratz EA. Oral alpha-tocopherol supplements decrease plasma gamma-tocopherol levels in humans. *J Nutr* 1985;115:807–13.
- (21) Fleshner NE, Klotz LH. Diet, androgens, oxidative stress and prostate cancer susceptibility. *Cancer Metastasis Rev* 1998;17:325–30.
- (22) Ricciarelli R, Zingg JM, Azzi A. Vitamin E: protective role of a Janus molecule. *FASEB J* 2001;15:2314–25.
- (23) Zhang Y, Ni J, Messing EM, Chang E, Yang CR, Yeh S. Vitamin E succinate inhibits the function of androgen receptor and the expression of prostate-specific antigen in prostate cancer cells. *Proc Natl Acad Sci U S A* 2002;99:7408–13.
- (24) Ni J, Chen M, Zhang Y, Li R, Huang J, Yeh S. Vitamin E succinate inhibits human prostate cancer cell growth via modulating cell cycle regulatory machinery. *Biochem Biophys Res Commun* 2003;300:357–63.
- (25) Hartman TJ, Dorgan JF, Woodson K, Virtamo J, Tangrea JA, Heinonen OP, et al. Effects of long-term alpha-tocopherol supplementation on serum hormones in older men. *Prostate* 2001;46:33–8.
- (26) Traber MG. Vitamin E. In: Shils ME, Olson JA, Shike M, Ross AC, editors. *Modern nutrition in health and disease*. 9th ed. Baltimore (MD): Lippincott Williams & Wilkins;1999. p. 347–62.
- (27) Klein EA, Thompson IM, Lippman SM, Goodman PJ, Albanes D, Taylor PR, et al. SELECT: the next prostate cancer prevention trial. *J Urol* 2001;166:1311–5.

NOTES

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